

Bioavailability Study of Fixed-Dose Tablet Versus Capsule Formulation of Amlodipine Plus Benazepril: A Randomized, Single-Dose, Two-Sequence, Two-Period, Open-Label, Crossover Study in Healthy Volunteers

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ABSTRACT

Background: In the treatment of hypertension, combination therapy is important because antihypertensive monotherapy is effective in only 40% of patients worldwide. Amlodipine is a dihydropyridine calcium channel blocker with a slow onset and long duration of action. Benazepril hydrochloride is a prodrug hydrolyzed by esterase to the active metabolite benazeprilat, an angiotensin-converting enzyme inhibitor. In 1995, the US Food and Drug Administration approved the use of a capsule formulation of combination amlodipine-benazepril for hypertension.

Objective: The aim of this study was to compare the bioavailability and tolerability of the capsule formulation with those of a tablet formulation of combination amlodipine-benazepril in healthy volunteers.

Methods: This single-dose, 2-sequence, 2-period, open-label, crossover study recruited healthy, adult, male volunteers with normotension. Subjects were randomly assigned to 1 of 2 treatment sequences: a single-dose tablet containing amlodipine 5 mg plus benazepril 10 mg, followed by a single-dose capsule containing the same dose of each drug (AB), or vice versa (BA). The treatment period for each drug consisted of dosing and pharmacokinetic analysis on day 1, followed by pharmacokinetic analysis on days 2 to 7. Treatment periods were separated by a 4-week washout period. For pharmacokinetic analysis, serial blood samples were obtained before dosing and at 20, 40, 60, 80, and 100 minutes and 2, 3, 4, 5, 6, 7, 8, 10, 12, 24, 36, 60, 84, 108, 132, and 156 hours after dosing. Tolerability was assessed using subject interview and spontaneous reporting.

Results: Twelve healthy, male, Taiwanese subjects (mean [SD] age, 23.5 [1.7] years) participated in the study. No statistically significant differences in bioavailability were found between the 2 formulations based on the pharmacokinetic measurements of amlodipine and benazeprilat. The rate and extent of

absorption of the tablets were found to be comparable to those of the capsules (90% CI, between 80% and 125%). The mean (SD) relative bioavailabilities, as represented by $AUC_{0-\infty}$, of amlodipine and benazeprilat for tablets versus capsules were 1.060 (0.170) versus 0.949 (0.197), respectively. The mean plasma concentration–time profiles of amlodipine and benazeprilat were graphically similar. No adverse effects were observed with either formulation.

Conclusions: The results of this bioavailability comparison study in this population of healthy, male, Taiwanese volunteers suggest that the tablet and capsule formulations of combination amlodipine-benazepril are bioequivalent. Both formulations were well tolerated. (*Curr Ther Res Clin Exp.* 2005;66:69–79) Copyright © 2005 Excerpta Medica, Inc.

Key words: bioequivalence, bioavailability, pharmacokinetics, amlodipine besylate, benazepril hydrochloride, fixed-dose combination.

INTRODUCTION

In the treatment of hypertension, combination therapy is important because antihypertensive monotherapy is effective in only 40% of patients worldwide.¹ Products containing a combination of 2 classes of antihypertensive drugs (eg, a calcium channel blocker [CCB] and an angiotensin-converting enzyme inhibitor [ACEI]) result in synergistic effects on blood pressure control and vital-organ protection, and decrease the risk for adverse effects (AEs). The Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure² also recommends combination therapy.

Among various combinations, those containing a CCB plus an ACEI are prescribed most often.^{3–5} Long-acting CCB-ACEI combination therapy (eg, amlodipine-benazepril) is considered effective in treating congestive heart failure and providing renal protection in patients with hypertension.³

Amlodipine, a dihydropyridine CCB with a slow onset and long duration of action, can inhibit the influx of extracellular calcium across vascular smooth-muscle cell membranes. The resulting decrease in intracellular calcium inhibits the contractile processes of myocardial smooth-muscle cells, resulting in dilation of the coronary and systemic arteries.⁶ Benazepril hydrochloride is a prodrug hydrolyzed by esterase to the active metabolite benazeprilat. The latter has a long duration of action and decreases blood pressure by inhibiting angiotensin II production.⁷ A capsule formulation* containing a combination of amlodipine 5 mg plus benazepril 10 mg was approved in March 1995 by the US Food and Drug Administration for the treatment of hypertension. Compared with amlodipine and benazepril monotherapy at the same doses, this combination is associated with a lower risk for edema and a greater decrease in blood pressure.⁸

*Trademark: Lotrel® (Novartis Pharmaceuticals Corporation, East Hanover, New Jersey).

Tablet and capsule formulations are both commercially available. A tablet formulation* might have pharmacokinetic properties different from those of the capsule formulation. The tablet is scored and thus relatively easy to separate, if needed. If the relative bioavailability and pharmacokinetic properties are comparable between the 2 formulations, pharmaceutical manufacturers would have an additional choice in generic drug development, and physicians could choose which formulation would be most appropriate in individual patients. However, the bioavailabilities of differing formulations may vary.

The purpose of this study was to compare the relative bioavailability (ie, the rate and extent of absorption) and tolerability of the tablet versus the capsule formulation of combination amlodipine-benazepril in healthy volunteers.

SUBJECTS AND METHODS

This study was conducted from May 5, 2002, to June 8, 2002, at the Clinical Trial Center of the National Taiwan University Hospital, Taipei, Taiwan. The study protocol was reviewed and approved by the institutional review board (IRB) at the hospital. The IRB was to be informed of any serious or unexpected AEs that might affect the safety of the subjects or the conduct of the trial. All experiments complied with the Good Clinical Practice guidelines.

Inclusion and Exclusion Criteria

Healthy male volunteers aged ≥ 18 years with normotension were recruited from the general population.

Subjects were excluded from the study if findings on physical examination, biochemistry (blood urea nitrogen, serum creatinine concentration), urinalysis, or hematology were abnormal. Patients were excluded if they had received any other medications within 14 days before the study. We did not perform pharmacogenomic analyses of metabolic rates/differences or determine acetylator status.

All subjects provided written informed consent, underwent complete screening including laboratory analysis, and were randomized 1 week before the start of the study.

Study Drug Administration

A single-dose, 2-sequence, 2-period, open-label, crossover design was used. Subjects were randomly assigned, using a computer-generated list of random numbers, to 1 of 2 treatment sequences: a single-dose tablet containing amlodipine 5 mg plus benazepril 10 mg, followed by a single-dose capsule containing the same dose of each drug (AB), or vice versa (BA). Both treatments were to be received after a 10-hour overnight fast. The treatment period for each drug consisted of dosing and pharmacokinetic analysis on day 1, followed by pharma-

*Trademark: Latrel, Amtrel® (TTY Biopharm Co., Ltd., Taipei, Taiwan).

cokinetic analysis on days 2 to 7. The 2 treatment periods were separated by a 4-week washout period.

Nicotine, alcohol, and caffeine use was not permitted for at least 48 hours before and during the study. Other medications and strenuous exercise were not allowed during the study. On the day of study drug administration in both treatment periods, lunch and dinner were provided to all subjects at the same times of day and were of similar caloric and fat content and distribution. Qualified health care professionals attended to the subjects throughout the study.

Laboratory Analysis

For the purposes of serial blood sampling for pharmacokinetic analysis, subjects remained at the hospital for 12 hours after dosing, and returned to the hospital on the morning and evening of study day 2 and on the evenings of days 3 to 7. Specifically, blood samples were obtained before dosing and at 20, 40, 60, 80, and 100 minutes and 2, 3, 4, 5, 6, 7, 8, 10, 12, 24, 36, 60, 84, 108, 132, and 156 hours after dosing. The samples were drawn by a nurse using a needle stick or IV cannula. If a cannula was used, it was inserted into an arm vein within 5 minutes before dosing and was maintained using isotonic saline for flushing. Immediately after samples were obtained, they were centrifuged at 4°C at ~3000 rpm for 10 minutes, and the plasma was transferred to appropriately labeled polypropylene tubes. Samples were frozen in an upright position at -20°C and stored at this temperature until shipment to a central laboratory (Protech Pharmservices Corporation, Taipei, Taiwan) for assay.

Assessment of AEs on each study day included subject interview, spontaneous reporting, laboratory analysis, physical examination, and electrocardiography.

A sensitive, specific, accurate, reproducible liquid chromatography/mass spectrometry/mass spectrometry (LC/MS/MS) method was used to determine plasma amlodipine and benazeprilat concentrations.⁹ Recovery studies were conducted to validate the analytical procedure based on the primary performance characteristics (precision, accuracy [including within-run and between-run variation], linearity, specificity, reproducibility, and limit of quantification). The lower limits of quantification of amlodipine and benazeprilat achieved were 0.1 and 0.2 ng/mL, respectively. Recovery and reproducibility assessment indicated good precision. Peak area ratios were used for calculation, and the calibration curve was fitted to a weighted ($1/x$) linear regression model; the linearity of this procedure was indicated by an average correlation coefficient of ≥ 0.998 . The coefficient of variation (CV) of concentrations in the calibration curves ranged from 1.0% to 8.9% in the plasma samples. The relative errors of the concentrations in the calibration curves ranged from -5.2% to 9.5%.

Pharmacokinetic Analysis

Plasma concentration-time data for amlodipine and benazeprilat were tabulated and graphically displayed for each subject (data not shown). These data are presented as mean (SD). Based on these data, C_{\max} , T_{\max} , AUC_{0-t} , $AUC_{0-\infty}$, $t_{1/2}$,

and the elimination rate constant (k_e) were calculated using standard noncompartmental analytical methods. The k_e value was determined using simple linear regression based on the terminal phase of plasma concentration. AUC_{0-t} was determined using the trapezoidal rule. $AUC_{0-\infty}$ was determined using the trapezoidal rule and extrapolated to infinity using an estimate of the last quantifiable concentration divided by k_e . The $t_{1/2}$ value was estimated using $(0.693/k_e)$. $AUC_{0-\infty}$ at (first) moment ($AUMC_{0-\infty}$) was determined using the trapezoidal rule and extrapolated to infinity using the following equation:

$$AUMC_{0-\infty} = C_n t_n / k_e + C_n / k_e^2 + \sum [(t_n - t_{n-1}) \times (C_{n-1} t_{n-1} + C_n t_n) / 2],$$

where C_n is the plasma drug concentration, t_n is the time point, t_{n-1} is the previous time point, and C_{n-1} is the plasma drug concentration at the previous time point.

Mean residence time (MRT) was determined using the following equation:

$$MRT = AUMC_{0-\infty} / AUC_{0-\infty}$$

The comparative bioavailability of the tablet and capsule formulations was summarized using the relative rate of absorption, comparing the amlodipine and benazeprilat C_{max} values and MRTs and the ratio of the AUC.

Statistical Analysis

Given a 20% bioequivalence limit, a 3% difference in concentration-time data between the 2 formulations, and an estimated CV of 15%, 12 subjects would be required to achieve an 80% power at the 5% nominal level, based on Schuirmann's 2 one-sided test procedures using a 2×2 crossover design.⁹

For each of the derived parameters, summary statistics (n , mean, median, SD, minimum, maximum, and CV) were calculated. Comparisons between the 2 formulations were made using the mean of the raw data of these parameters. For AUC and C_{max} comparisons, we used log-transformed (\ln) data due to a skewed distribution. For MRT and $t_{1/2}$, the means of the raw data were compared using the Student t test. Continuous variables were analyzed using analysis of variance for the crossover design. The Wilcoxon Mann-Whitney test was used for the nonparametric method if the distribution was not normal, such as that of T_{max} . The 90% CI between the 2 dosing formulations was assessed. Based on the plasma concentration-time data, the pharmacokinetic parameters were determined with noncompartmental methods using WinNonlin Professional version 3.1 (Pharsight Corporation, Palo Alto, California).

RESULTS

Twelve healthy, male, Taiwanese subjects participated in the study (mean [SD] age, 23.5 [1.7] years [range, 21–27 years]; mean [SD] body weight, 65.7 [8.5] kg

[range, 57–73 kg]; mean [SD] height, 171.4 [4.6] cm [range, 157.0–177.8 cm]; mean [SD] body mass index, 22.39 [2.25] kg/m² [range, 18.25–25.96 kg/m²].

No statistically significant differences in C_{\max} , T_{\max} , $\ln(\text{AUC}_{0-t})$, $\ln(\text{AUC}_{0-\infty})$, MRT, or $t_{1/2}$ were found between the 2 formulations (**Table I**). The 90% CIs for the rate and extent of absorption ($\ln[\text{AUC}_{0-t}]$, $\ln[\text{AUC}_{0-\infty}]$, and $\ln[C_{\max}]$) of the tablet versus the capsule were between 80% and 125%. The mean (SD) relative bioavailabilities, as represented by $\text{AUC}_{0-\infty}$, of amlodipine and benazeprilat for tablets versus capsules were 1.060 (0.170) versus 0.949 (0.197), respectively. The mean plasma concentration–time profiles of amlodipine and benazeprilat were graphically similar (**Figure**).

No clinically significant changes in vital signs, physical examination findings, laboratory parameters, or electrocardiography were found. No clinical AEs were observed.

DISCUSSION

This comparative bioavailability study suggests that single-dose tablets and capsules of combination amlodipine–benazepril had statistically similar pharmacokinetic properties in healthy, male, Taiwanese subjects. No statistically significant differences in C_{\max} or T_{\max} were found between the 2 formulations, suggesting that the rate of absorption of the tablets was similar to that of the capsules. The 90% CIs for the rate and extent of absorption ($\ln[\text{AUC}_{0-t}]$, $\ln[\text{AUC}_{0-\infty}]$, and $\ln[C_{\max}]$) of the tablets versus the capsules were between 80% and 125%, suggesting bioequivalence. No significant differences in MRT or $t_{1/2}$ were found between the 2 formulations, indicating that the retention time and elimination rate were statistically similar between them. Moreover, no clinical AEs were observed during the study.

The values for the pharmacokinetic properties obtained from the healthy volunteers in the present study were comparable to those reported previously for amlodipine and benazeprilat (**Table II**).^{10–12} The T_{\max} and $t_{1/2}$ of amlodipine achieved with a single capsule in the present study were similar to findings in previous studies, indicating that the absorption and elimination rates in the present study were similar to those achieved in previous studies. In our study of amlodipine 5 mg and benazepril 10 mg, the amlodipine bioavailability measures were half of those found in the study by Faulkner et al,¹⁰ in which amlodipine 10 mg was administered to white male volunteers. Consequently, it might be concluded that the bioavailability of amlodipine is similar in Taiwanese compared with white volunteers.

The C_{\max} , T_{\max} , and AUC_{0-t} of benazeprilat achieved with the capsule in the present study were similar to those found in previous studies.^{11,12} However, significant differences in $t_{1/2}$ were found between 3 studies (all, $P < 0.05$), with the highest values occurring in the study by Kaiser et al.¹² Thus, it could be postulated that the longer the sampling time, the more compartments could be seen in the profile of benazeprilat, which was used to calculate the $t_{1/2}$. In the study by Kaiser et al,¹² the mean sampling time was 22.3 hours.

Table 1. Pharmacokinetic parameters of amlodipine and benazeprilat* after single dosing with a tablet and capsule formulation of combination amlodipine 5 mg plus benazepril 10 mg in healthy volunteers.

Parameter	Amlodipine				Benazeprilat			
	Tablet, Mean (SD)	Capsule, Mean (SD)	90% CI	P	Tablet, Mean (SD)	Capsule, Mean (SD)	90% CI	P
AUC ₀₋₁₂ ng/mL·h	125 (33)	122 (46)	97.1–116.0	0.256	1403 (256)	1528 (370)	83.2–104.0	0.246
AUC _{0-∞} ng/mL·h	136 (39)	134 (53)	96.5–113.0	0.339	1464 (253)	1593 (377)	82.9–104.0	0.276
C _{max} ng/mL	2.92 (0.61)	2.92 (0.95)	93.1–114.0	0.618	239 (49)	255 (57)	83.9–105.0	0.347
T _{max} h	5.67 (0.89)	6.33 (1.92)	–	–	1.78 (0.64)	1.86 (0.44)	–	–
MRT, h	58.1 (9.6)	59.0 (10.5)	94.4–103.0	0.530	13.0 (6.4)	12.8 (7.6)	76.8–128.0	0.878
t _{1/2} h	41.1 (7.2)	42.3 (7.3)	92.8–102.0	0.264	17.9 (12.7)	17.1 (16.60)	63.5–146.0	0.840
F score	1.060 (0.170)		95.1–116.0	0.258	0.949 (0.197)		82.4–107.0	0.391

MRT = mean residence time.
*Benazeprilat is the active metabolite of benazepril.

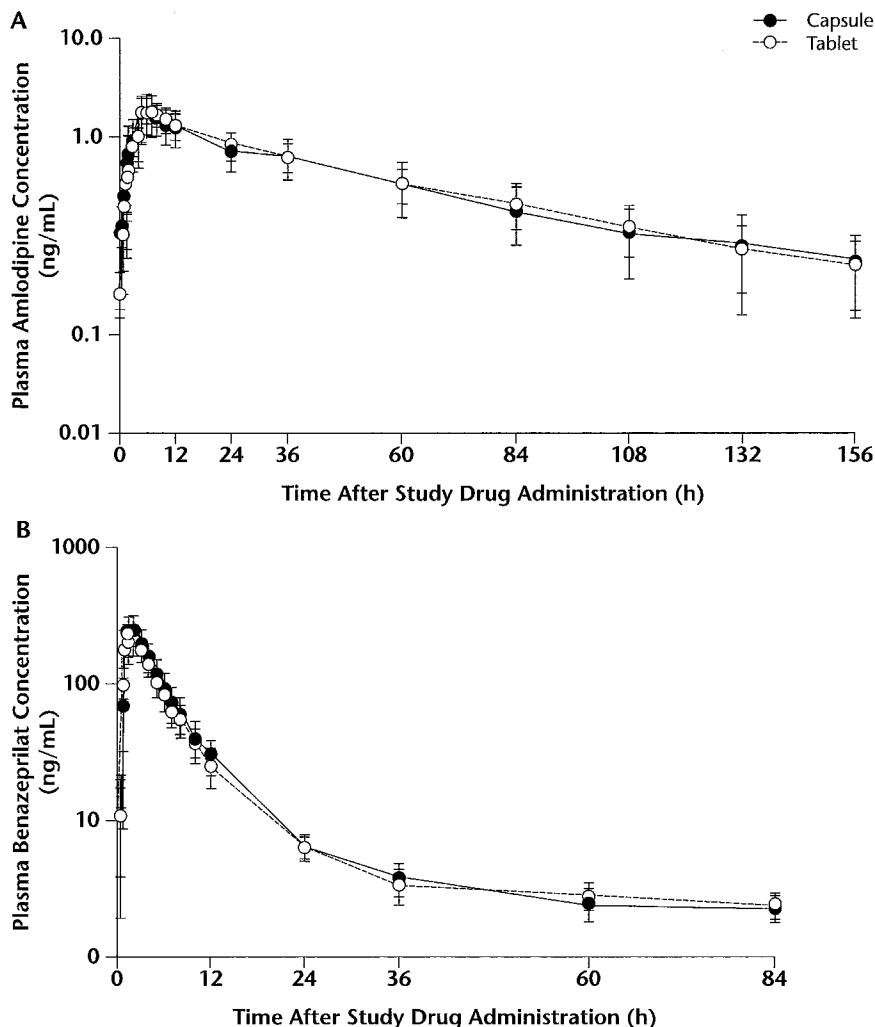


Figure. Mean (SE) plasma concentrations of (A) amlodipine and (B) benazeprilat (the active metabolite of benazepril) before (time 0; baseline) and after single dosing with a tablet and capsule formulation of combination amlodipine 5 mg plus benazepril 10 mg in healthy volunteers.

Oral amlodipine is slow acting but is almost completely absorbed in the gastrointestinal tract, and in one study, the absolute bioavailability after oral administration was found to be relatively high in healthy volunteers (mean C_{max} , ≤ 5.9 ng/mL).¹⁰ In another study, AUC, C_{max} , and T_{max} were consistently stable.¹³ Due to extensive distribution and relatively slow clearance, amlodipine has been found to have a long $t_{1/2}$, ranging from 31 to 50 hours.^{10,13,14} Benazepril has been found to be absorbed from the gastrointestinal tract and

Table II. Pharmacokinetic parameters of amlodipine and benazeprilat* in the present study compared with previous studies.

Study/Treatment	Dose, mg	Formulation	C _{max} [†] Mean (SD), ng/mL	T _{max} [†] Mean (SD), h	t _{1/2} [†] Mean (SD), h	AUC _{0-∞} [†] Mean (SD), ng/mL · h
Amlodipine						
This study (reference drug)						
Amlodipine + benazepril [†]	5	Capsule	2.92 (0.95)	6.3 (1.9)	42.3 (7.3)	122 (46)
Faulkner et al ¹⁰						
Amlodipine monotherapy	10	Capsule	5.9 (1.2)	7.6 (1.8)	35.7 (6.1)	238 (53)
Sun et al ¹¹						
Amlodipine monotherapy	5	Tablet	2.3 (0.6)	9.0 (2.6)	36.3 (6.5)	114 (54)
Amlodipine + benazepril [‡]	5	Tablet	2.5 (0.8)	8.3 (2.2)	36.3 (6.5)	118 (53)
Benazeprilat						
This study (reference drug)						
Amlodipine + benazepril [†]	10	Capsule	255 (57)	1.9 (0.4)	17.1 (16.6)	1528 (370)
Sun et al ¹¹						
Benazepril monotherapy	10	Tablet	260 (93)	1.5 (0.6)	5.6 (3.2)	1410 (384)
Amlodipine + benazepril [‡]	10	Tablet	292 (101)	1.7 (0.7)	5.6 (3.2)	1470 (358)
Kaiser et al ¹²						
Benazepril monotherapy	10	Capsule	195 (54)	1.5 (0.4)	22.3 (9.2)	1261 (272)

*Benazeprilat is the active metabolite of benazepril.

[†]Amlodipine and benazepril were administered as 1 combination capsule.[‡]One amlodipine tablet and 1 benazepril tablet were coadministered.

then converted to benazeprilat.¹⁵ The mechanisms of action have been shown to be mainly via hepatic metabolism, and conversion to be virtually complete after 4 hours, with consistently stable pharmacokinetic properties.¹²

One previous clinical trial¹⁶ assessed the tolerability of the oral combination amlodipine-benazepril in >1600 patients with hypertension; >500 of these patients were treated for ≥ 6 months and >400 were treated for >1 year. The reported AEs were generally mild and transient, and no correlation with age, sex, race, or duration of therapy was found. In the present study, no AEs were observed, perhaps due to the small sample size.

Sun et al¹¹ found that the rate and extent of absorption of amlodipine and benazepril in oral combination therapy were not statistically different from those of either drug used alone. Sun et al¹¹ also studied the pharmacokinetic interaction between amlodipine and benazepril in 12 healthy male subjects. Single doses of amlodipine 5 mg and benazepril 10 mg were orally administered alone or in combination according to a 3-way, Latin square, randomized, cross-over design. Our results, echoing those of Sun et al,¹¹ indicated no pharmacokinetic interaction between the 2 drugs.

CONCLUSIONS

The results of this bioavailability comparison study in this population of healthy, male, Taiwanese volunteers suggest that the tablet and capsule formulations of combination amlodipine-benazepril are bioequivalent. Both formulations were well tolerated.

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